REMARKS

The Applicants respectfully request reconsideration of claims 1 – 9, 11-17, 36, and 48 – 56, in light of the amendments and arguments submitted herewith. The Applicants also requests consideration of new claims 57 and 58.

Cancelled Claims and Amendment to Claim 56

Claims 10, 22, and 23 are cancelled without prejudice, the Applicants reserving the right to prosecute such claims in a follow-on application. Claim 56 is amended to correct a typographical error, replacing the word "method" with the phrase "reagent cell population" as is evident from the context of the claim.

Indefiniteness

Claim 48 is amended to delete the term "foreign" from the claim. As such, the claim specifically delineates what is meant by "genetic material," and complies with the requirements of 35 U.S.C. §112, second paragraph.

Prior Art Rejections

Pending claims 1-4, 6, 36, and 48-56 stand rejected as being anticipated by Smith et al. (U.S. Patent No. 6,146,888). As well, claims 1-9, 11-17, 36, and 48-56 stand rejected as being obvious in light of some combination of Smith, Myers (Molecular Biology and Biotechnology, ed. Myers, VCH Publishers, Inc., 1995, pp. 165 - 168); Fasbender et al. (Jour. Biol. Chem., 272(10):6479 – 6489, 1997); Pascolo et al. (J. Exp. Med., 185:2043 – 2051, 1997); Thomson (Science, 282:1145 – 1147, 1998); and Bradley et al. (U.S. Patent No. 5,614,396). In light of amendments to claims 1 and 11, and the foregoing arguments, all pending claims are patentable.

A. Claims 1-9, 11-17 and New Claims 57 and 58

Claims 1 and 11 are amended to include limitations regarding the efficiency of genetic material introduction. In particular, claim 1 is amended to include the limitation that "the polynucleotide is introduced . . . by transfection in the presence of a chemical

agent" and that the "transfection efficiency . . . exceeds the efficiency for transfection human embryonic stem cells using electroporation . . ." Claim 11 is amended to include the limitation that "the efficiency of introducing the DNA sequence exceeds the efficiency for transfecting the DNA sequence into human embryonic stem cells using electroporation." Support for the amendments may be found in the results shown in Figure 1 of the application and at page 12, lines 1-19.

With these amendments, claims 1-9 and 11-17 are patentable because none of the prior art, alone or in any combination, teaches or suggests a method of introducing a polynucleotide into a human embryonic stem cell with a transfection efficiency exceeding the efficiency of using electroporation to introduced the polynucleotide into the human embryonic stem cell.

1. Anticipation

Smith teaches the use of electroporation to transfect murine embryonic stem cells. Smith does not teach the use of a chemical agent for transfection, or the superiority of a chemical agent in transfecting a polynucleotide into a human embryonic stem cell. As well, Smith in no way enables the transfection of human embryonic stem cells at any level because not a single experiment is described using *human* embryonic stem cells. Thus Smith cannot anticipate claim 1. For similar reasons, Smith does not anticipate claim 11 since the reference does not teach the use of cationic polymers to introduce DNA into a human embryonic stem cell, and does not teach the superiority of the method over electroporation. Since claims 2-9 and 12-17 depend from claims 1 and 11, respectively, all of claims 1-9 and 11-17 are not anticipated by Smith.

2. Nonobviousness

Furthermore claims 1-9 and 11-17 are not obvious in light of any combination of the cited art because none of the cited art teaches the introduction of polynucleotide, or DNA, into a human embryonic stem cell with an efficiency that exceeds that for electroporation.

As previously discussed, Smith only teaches the transfection of DNA into murine embryonic stem cells using electroporation.

Fasbender is drawn to using a cationic polymer, or a cationic lipid, with an adenovirus to infect human epithelia or nasal epithelium of cystic fibrosis in mice (see abstract). Fasbender does not teach (i) the use of any of its techniques as applied to human *embryonic stem cells* and (ii) that the use of chemical agent results in improved transfection efficiency in transferring polynucleotide, or DNA, to a human embryonic stem cell over electroporation, as required by claims 1 and 11.

Thus a combination of Smith and Fasbender cannot provide a basis for a prima facie case of obviousness because neither reference teaches the necessary elements of (i) introducing a polynucleotide, or DNA, in the presence of a chemical agent with an efficiency that exceeds that of electroporation; and (ii) transfecting human embryonic stem cells; as required by claims 1 and 11.

Independent claim 1 is amended to specifically require that introduction of the polynucleotide result in a "transfection efficiency that exceeds the efficiency for transfecting human embryonic stem cells using electroporation." This unexpected, superior result is in no way taught, suggested, or motivated by any of the cited art.

Furthermore, neither Smith nor Fasbender specifically addresses transfection of *human embryonic stem cells*; one cannot simply combine their teachings to obtain the invention in claims 1 and 11 with a reasonable expectation of success. For example, Smith teaches the use of electroporation to transfect murine embryonic stem cells. The present application shows the superiority of chemical agents over electroporation (see Figure 1 of the application), which underscores the difference between human and murine embryonic stem cells.

Additionally, as pointed out in the previous response, Fasbender shows that Lipofectamine works extremely well in transfecting fliosarcoma cells (see Figure 4 of Fasbender), while the present application shows Lipofectamine to work extremely poorly in transfection human embryonic stem cells (see Figure 1 of the present application). As well, Fasbender states "[n]onviral cationic vectors . . . do not catalyze the subsequent steps in gene transfer" (see Fasbender abstract), while the present application shows such vectors have surprising efficiency in gene transfer versus electroporation. Thus embryonic stem cells cannot be reasonably expected to transfect in the same manner as other types of cells.

The Office Action mentions that Applicants' previous response states that "an hES cell could be transfected, even with poor efficiency" (see Office Action, page 10, first paragraph). Such a statement was only made in the context of electroporation (see Applicants' response of December 19, 2003, page 15, first full paragraph), and has no bearing at all with regard to a combination of Smith and Fasbender to transfect a hES in the presence of a chemical agent.

Therefore, one skilled in the art cannot combine Smith and Fasbender a priori to assume a reasonable expectation of success in transfecting human embryonic stem cells. Embryonic stem cells do not act like all other cells under transfection. Human embryonic stem cells are different from murine embryonic stem cells.

Myers and Pascolo are directed toward the teaching of specific fluorescent proteins and knockout genomic sequences, but do not provide any teaching about transfection of human embryonic stem cells, or the improved efficiency of transfection of human embryonic stem cells in the presence of chemical agents relative to electroporation. Thus Myers and Pascolo, in combination with any of the other cited art, cannot render claims 1 and 11 obvious.

Finally, neither Thomson or Bradley teaches claims 1 and 11. Thomson teaches the harvesting of human ES cells from the inner cell masses of blastocysts and Bradley et al. teach the transfection of embryonic stem cells using electroporation and homologous recombination. Though Bradley et al. state that the invention may be performed with human cells, the data presented only show homologous recombination using the AB1 cell line, a murine embryonic cell line (see Examples in cols. 29 – 36). Neither reference teaches the use of a chemical agent for transfection of human embryonic stem cells, or that the chemical agent has a greater transfection efficiency than electroporation. Thus the combination of the two references fails to make a prima facie case of obviousness.

Thus, claims 1 and 11 are not obvious in light of any combination of the cited art. Since claims 2-9 and 12-17 all depend from nonobvious claims 1 and 11, respectively, all of claims 1-9 and 11-17 are not obvious. Therefore, claims 1-9 and 11-17 are patentable.

3. New Claims

Finally new claims 57 and 58, dependent from claims 1 and 11 respectively, are added to include the limitation of an absence of an adenovirus. Support for such a limitation may be found in the application (see page 4, line 24; and Example 1 on page 18 that describes a transfection protocol without the use of an adenovirus). Claims 57 and 58 are patentable over the cited art since none of the art suggests or motivates, alone or in combination, the transfection of human embryonic stem cells using a chemical agent in the absence of an adenovirus.

B. Claims 36 and 48-56

In order to expedite prosecution, claims 36 and 48 are amended to include the limitation that the cell population (claim 36) or reagent cell population (claim 48) is produced in accordance with claim 1. Thus claims 36 and 48 are patentable for substantially the same reasons as claim 1. Claims 49-56, being dependent from claim 48, are thus also patentable over the cited art. The Applicants maintain, however, that former claims 36 and 48-56 are still patentable. The Applicants reserve the right to prosecute the former form of claims 36 and 48-56 in a related continuation application.

Conclusion

In view of the amendments and arguments presented, the Applicants respectfully request allowance of pending claims 1-9, 11-17, 36, and 48 – 58. The representative of the Applicants cordially requests a telephone conference with the Examiner to discuss this paper, if such communication would help expedite the prosecution of the application.

Respectfully submitted,

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